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(71) Applicant: MALLINCKRODT MEDICAL, INC. [US/US]; 675 McDonnell Blvd., P.O. Box 5840, St. Louis, MO 63134 (US).

(72) Inventor: RAJAGOPALAN, Raghavan ; 12031 Vinson Court, Maryland Heights, MO 63043 (US).

(74) Agents: VACCA, Rita, D. et al.; Mallinckrodt Medical, Inc., 675 McDonnell Boulevard, P.O. Box 5840, St. Louis, MO 63134 (US).

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(54) Title: TISSUE SPECIFIC IMAGING AGENTS USING INTERNAL IMAGE ANTI-IDIOTYPIC ANTIBODIES

(57) Abstract

A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, or recombinant anti-idiotypic antibody labeled with a chelate capable of intravenous injection into an animal to produce reliable visual imaging of biological receptors.

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TISSUE SPECIFIC IMAGING AGENTS USING INTERNAL IMAGE ANTI-IDIOTYPIC ANTIBODIES

Field of the Invention

The present invention relates generally to the use of labeled anti-idiotypic antibodies for diagnostic imaging and, more particularly, to labeled anti-idiotypic antibodies for use as agents to image in vivo receptors in biological systems for diagnostic use.

Background of the Invention

The idiotypic network theory of Jerne (Jerne, N.K., *Anals. Inst. Pasteur Paris* 125c, 372) proposes that the variable regions of antibodies (i.e. idiotypes) act as immunogens to give rise to a secondary set of antibodies called "anti-idiotypes". In particular, if antibodies are developed against a ligand that binds to a certain receptor within the body, then the resulting anti-idiotypic population may contain antibodies that will likewise bind to the same receptor due to each the ligand and the anti-idiotypic antibody having similar topological features. Essentially the anti-idiotypic antibody mimics the ligand. Application of the principles proposed by Jerne has led to the development of a number of anti-idiotypic antibodies such as those against acetylcholine, TSH, glucocorticoid, adenosine and similar such compounds, without ever having to isolate and purify the natural receptor (Erlanger, B.F., *Inter. Rev. Immunol.*, 5, 1989, 131) which can be quite difficult.

The use of radiographic imaging agents for the visualization of skeletal structures, organs, or tissues is also well known in the area of biological and medical research and diagnostic procedures. The procedure whereby

such imaging is accomplished generally involves the preparation of radioactive agents, which, when introduced to the biological subject, are localized in the specific skeletal structures, organs, or tissues to be studied. The localized radioactive agents may then be traced, plotted, or scintiphotographed by radiation detectors such as traversing scanners or scintillation cameras. The distribution and relative intensity of the detected radiation indicates the position of the agent in the tissue and also shows the presence of aberrations, pathological conditions and the like. The density and distribution of the receptors being so imaged, depends on the pathological state of that particular tissue.

The specific targeting of effector molecules to a particular tissue, such as a tumor, using monoclonal antibodies is also well known in the art (Halpern, S.E., et al., *Diagnostic Imaging*, 1983, 40). Recently, technetium-99m or indium-III labeled anti-myosin antibodies have been used to image myocardial infarction (Dean, R.T., et al., *J. Nucl. Med.*, 1989, 30, 934). Each of these approaches to imaging particular tissue areas are based upon the ability of a particular type of cell to secrete a particular substance in a very high concentration compared to other cells in the vicinity of the desired area to be imaged.

Therefore, a need exists to provide an approach to site specific internal diagnostic imaging of tissue areas which does <u>not</u> necessitate particular cell types having the ability to secrete a substance in very high concentrations compared to other cells.

In general, it is an object of the present invention to provide an unique receptor mediated approach, as opposed to an approach dependent on the concentration of

secretions from various cells as described above, to image site specific areas of tissue. The particular anti-idiotypic antibodies of the present invention provide many advantages when used as diagnostic agents to provide a means of imaging biological receptors without having to isolate and purify the natural receptor.

Additional objects and features of the present invention will appear from the following description in which a preferred embodiment is described in detail.

Summary of the Invention

The present invention employs the use of anti-idiotypic antibodies for site specific diagnostic imaging of biological receptors without having to isolate and purify the natural receptor. An anti-idiotypic antibody refers to an antibody raised against a first antibody which specifically binds to the antibody binding site or CDR of the first antibody. The antibody binding site or CDR of an antibody is that portion thereof which specifically binds to the recognized epitope.

The anti-idiotypic antibodies of the present invention are made by developing antibodies against a first antibody that binds specifically to a certain desired ligand directed at the receptor within the body. The resulting anti-idiotypic antibody binds to the same receptor due to its topological similarity with the ligand.

The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment may be labeled with a radionuclide such as Tc-99m using a chelate approach wherein one of the preferred chelates is a multidentate organic compound with three amide nitrogen atoms and one

thiolate sulfur atom (N_3S chelate) bonded to a metal radionuclide, ^{99m}Tc, which is also bonded to one oxygen atom to form an anti-idiotypic antibody complex. The whole, fragmented or recombinant anti-idiotypic antibody or recombinant fragment thereof may likewise be labeled by fluorination or by complexing with a paramagnetic particle.

Following labeling, the anti-idiotypic antibody complex is then injected into a warm-blooded animal for site specific diagnostic imaging of the particular tissue area desired by means of imaging the labeled receptors thereof.

Detailed Description of the Invention

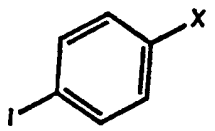
The anti-idiotypic antibody employed in the present invention is may be made according to the well established hybridoma technology as exemplified by European Patent Application Number 89103738.4, incorporated herein by reference.

The novel approach of utilizing the anti-idiotypic antibodies for imaging specific receptors within a desired tissue area has two major advantages over the conventional methods described above. First, it avoids having to use purified receptors to develop the anti-receptor antibodies. Often it is difficult and sometimes impossible to isolate pure, stable receptors for this type of immunization. Secondly, the attachment of a small molecule for labeling, e.g., molecules having a molecular weight of approximately 1000 of less, to a large anti-idiotypic antibody should not perturb its receptor binding capability significantly. In contrast, the classical bifunctional approach of attaching metal complexes directly to small effector substances, such

as drugs or hormones for example, essentially blocks the receptor binding capabilities.

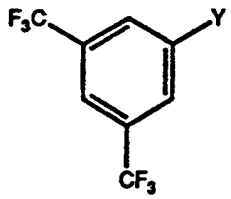
Diagnostically or therapeutically useful radionuclide elements which may be used to label the anti-idiotypic antibody include technetium, indium, rhenium, yttrium, gadolinium, gallium, bismuth, fluorine, iodine and the like which can be coupled to the whole, fragmented or recombinant anti-idiotypic antibodies or recombinant fragments thereof by any one of the several methods known in the art. Example methods that may be used in the present invention are disclosed in European Patent Application assigned publication number 0 284 071 and U.S. Patent Numbers 4,659,839; 4,732,974; 4,837,003 and 4,965,392, each incorporated herein by reference. As disclosed in these referenced patents, either the whole, fragmented or recombinant anti-idiotypic antibody or recombinant fragments thereof can be labeled with radionuclide chelates in a non-selective manner wherein the chelate is either bound at any location on the anti-idiotypic antibody or by a site-selective technique. In the site-selective technique, the radionuclide chelate is bound distally from the receptor binding site of the anti-idiotypic antibody by using, for example, a bifunctional coupling agent which reacts with a free sulfhydryl group generally found in the fragmented or anti-idiotypic antibody and is used to label the target biological receptors. A standard method for preparing anti-idiotypic antibody fragments is by the enzymatic digestion of the whole antibody with papain or pepsin as described by Parham, et al., *J. Immunol. Methods*, 1982, 53, 133. The anti-idiotypic antibody or fragments thereof can likewise be iodinated directly with a sodium iodide/chloramine-T procedure or can be attached via covalently bound bifunctional moieties such as those illustrated in Formula

I below:



FORMULA I

wherein X is selected from the group consisting of isocyanate, isothiocyanate, imidate, maleimido, succinimidyloxycarbonyl, acid chloride and sulfonyl chloride. Fluorines, which are potentially useful for fluorine magnetic resonance imaging (MRI) or for positron emission tomography (PET) can likewise, for example, be conjugated to the antibody via a bifunctional molecule as illustrated by Formula II below:



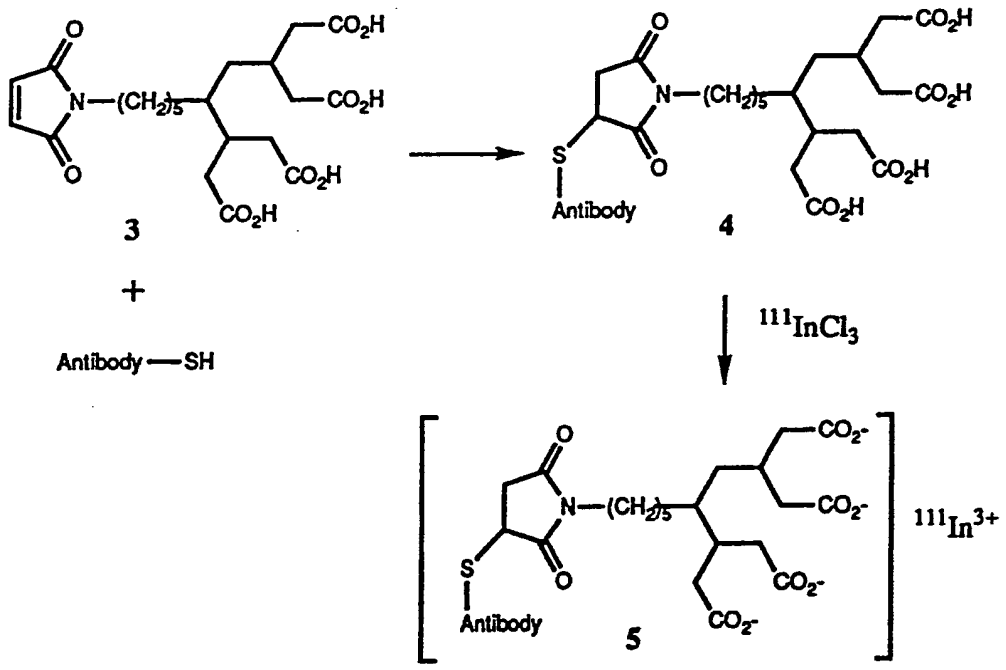
FORMULA II

wherein Y is defined to be the same as X above.

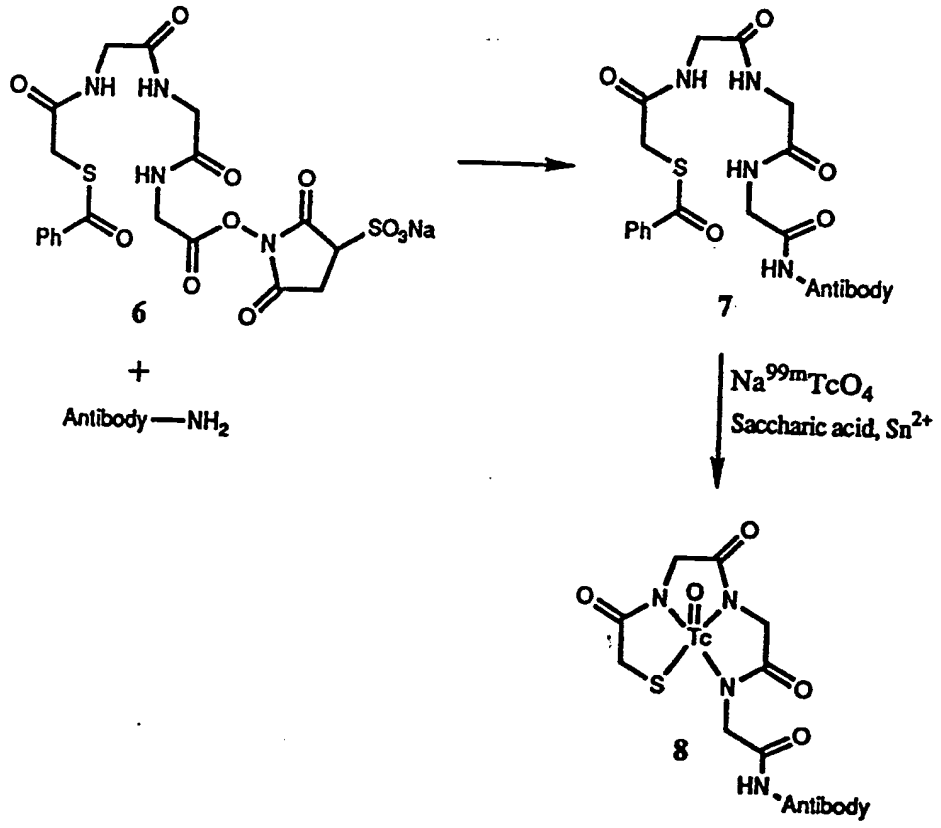
Metal ions such as technetium, rhenium, indium, yttrium, gadolinium, bismuth and the like can be joined to either the whole, fragmented or recombinant anti-idiotypic antibody or recombinant fragments thereof in a selective manner using a bifunctional molecule that contains an appropriate ligand and a coupling group that reacts specifically with the protein sulfhydryl groups such as a maleimido group as illustrated in Scheme I below. Alternatively, a non-selective manner may be utilized

wherein the bifunctional molecule contains the ligand and a coupling moiety selected from the group consisting of succinimidyloxycarbonyl, isocyanate, imidate, isothiocyanate, acid chloride and sulfonyl chloride such as illustrated in Scheme 2 below. The maleimido ligand 3, the succinimido ligand 6, and the method of labeling the conjugated proteins 4 and 7 with indium-111 or technetium-99m have been described in detail by Nicolotti, et al., U.S. Patent No. 4,732,974, incorporated herein by reference.

Scheme 1



Scheme 2



The present invention is therefore not restricted to radiographic imaging, and may be applied to any imaging modality. For example, the anti-idiotypic antibody of the present invention may be likewise labeled through fluorination or by labeling with a paramagnetic metal chelate. For further example, the antibody conjugate in Example 4 below can complex gadolinium or europium for MRI or immunofluorescence applications respectively.

In a preferred embodiment, the internal image antibody, i.e., anti-idiotypic antibody is directed at the digoxin receptor in the myocardium and is labeled with technetium-99m. It is believed that the labeled digoxin internal image antibodies may be useful in the diagnosis of some coronary disorders and may supplement the information gained from the use of myocardial perfusion agents such as thallium-201.

The novel imaging agents of this invention can be formulated into diagnostic compositions containing sufficient amount of labeled anti-idiotypic antibody for imaging, together with a pharmaceutically acceptable buffer such as phosphate, citrate, or tris(hydroxymethyl)amino-methane; balanced ionic solutions containing chloride and bicarbonate salts of blood plasma cations such as Ca^{2+}, N^{2+}, K^{+}, Mg^{2+}, saline and the like.

The concentration of the imaging agent according to the present invention should be sufficient to provide satisfactory imaging, c.a. 1 to 50 millicuries. The imaging agent should be administered so as to remain in the patient for 1 to 3 hours, although both longer and shorter time periods are acceptable. Therefore, convenient ampules containing 1 to 10 mL of aqueous solutions may be prepared.

Imaging may be carried out in the normal manner, for example by injecting a sufficient amount of the imaging composition to provide adequate imaging and then scanning with a suitable machine, such as a gamma camera.

The anti-idiotypic antibodies and the corresponding radionuclide conjugates can be prepared in accordance with the examples set forth below, which are not intended to be limiting.

EXAMPLE 1

Fusion of mouse myeloma cells with the spleen cells of AJ mice immunized with Balb-C mouse anti-digoxin antibody.

Monoclonal antibodies were produced by the hybridoma technology well known in the art. Two AJ mice were immunized with murine (Balb-C) monoclonal anti-digoxin antibody (Medex Laboratories). A booster injection was given 3 weeks after the primary immunization and the spleens were removed after 3 days. Mouse myeloma and the spleen cells were washed three times with Dulbecco's Eagle Medium (DME) and suspended in DME (10ml). A 5 mL portion of each of these cell suspensions were mixed and centrifuged. The supernatant was discarded and the pellet was treated with 1 mL of polyethylene glycol (added over a 45 second period), 3 mL of DME (added over 30 second period), and additional 9 mL of DME added over a 30 second period. The cells were allowed to stand at ambient temperature for 8 minutes and at 37° C for 2 minutes. The cells were centrifuged, suspended in HAT medium (10 mL), and distributed in microtiter plates. The cells were allowed to grow and were screened by radioimmunoassay procedure approximately 3 weeks after fusion.

EXAMPLE 2

Screening of anti-idiotypic antibodies for digoxin.

A 75μL portion of affinity purified goat anti-mouse antibody (2 mg/mL) was diluted with PBS buffer (150 mL). In each well was placed 200 μL of the above antibody and the plates were incubated at 37°C for 4 hours. The plates were washed with water (3 times), treated with 3% BSA solution (200 μL), and incubated for 1 hour. The wells were washed again with water (3 times) and then treated with the supernatants from the cell culture (150 μL) and allowed to incubate at ambient temperature for about 18 hours. The plates were washed with water (3 times), treated with ^{125}I labeled goat anti-digoxin (100 μL) and incubated for 4 hours. Thereafter, the plates were washed and the wells were counted. A total of 39 positive wells were identified.

The supernatants from the positive wells above (100 μL) were mixed with ^{125}I-digoxin (50 μL) and were placed in the microtiter plates which were previously coated with approximately 0.5 μg of monoclonal mouse anti-digoxin antibody for 1 hour. The plates were washed with water 3 times and counted. Inhibition of ^{125}I-digoxin, compared to the control, indicated a positive test for anti-idiotypes. Four positive wells were identified.

EXAMPLE 3

Preparation of $F_{ab'}$ fragment of anti-idiotypic digoxin antibody.

Ascites fluid is obtained in the usual manner by the injection of the hybridoma cells from Example 2 into mouse peritoneum. It is purified by three successive precipitation with ammonium sulfate using 20mM phosphate

buffer, pH 6.8. Thereafter, the protein is dialyzed exhaustively using 20 mM phosphate buffer, pH 6.8. The monoclonal antibody is purified by ion-exchange chromatography (Whatman C-52 column, 0 to 500 mM NaCl gradient in pH 6.8 phosphate buffer). The desired fraction is collected and stored in the same buffer at 4°C.

The desired amount of antibody (absorbance of 1% solution at 280 nm is 14.4) and cysteine-free papain (Worthington, 2 times crystallized) in the ratio of 1:20 are incubated at 37°C using 10 times the volume of digestion buffer (100 mM sodium acetate, 3 mM disodium EDTA, pH 5,5) until the reaction is complete (3-16 hours) as determined by SDS-PAGE. The digestion mixture is diafiltered (Amicon flow cell, PM-10 membrane) at 4°C using TRIS buffer, pH 7.2. It is then applied to the Whatman DE-52 ion-exchange resin, previously equilibrated in the same buffer, to remove the anionic F_c fragment. The eluent, which consists of $(F_{ab'})_2$ and inactivated papain, is purified by Sephadex G-100 size exclusion chromatography using TRIS buffer, pH 7.2. The desired antibody fragment elutes in the void volume and is characterized by SDS-PAGE. It is stored as frozen aliquots at -70°C.

The dimer thus obtained by papain digestion of the whole antibody is then further cleaved to the desired $F_{ab'}$ fragment using thiol reagents such as cysteine or dithiothreitol. The dimer in 25 mM phosphate buffer, pH 7.4, containing 2 mM disodium EDTA and 0.02% (w/v) sodium azide is incubated at room temperature with either cysteine or dithiothreitol until the reaction is complete as determined by SDS-PAGE (usually 1-4 hours). Excess reducing agent and other low molecular weight fragments are quickly removed by Sephadex G-25 column using PBS. The F_{ab} fragment thus obtained should be used as soon as possible

in order to prevent the oxidation of the sulfhydryl groups.

EXAMPLE 4

Site-selective labeling of $F_{ab'}$ fragments with indium-111.

A mixture of the $F_{ab'}$ fragment and about 20 fold excess of the ligand shown in Scheme 1 is incubated in labeling buffer (50 mM MES, pH 6.0) for 2-4 hours. Excess ligand and other low molecular weight impurities are quickly removed by Sephadex G-25 column using the labeling buffer.

The antibody fragment conjugated with the ligand is then labeled with radioactive indium chloride as described below. A mixture of $^{111}InCl_3$ (80 μL) and 4,5-dihydroxy-1,3-benzenedisulfonic acid (40 μL, 10 mM) in 0.2 M MES buffer (80 μL) is treated with the conjugated $F_{ab'}$ fragment and the entire mixture is incubated at room temperature for 1 hour. The reaction mixture is treated with 0.2 M EDTA (40 μL) to remove excess indium. The indium labeled antibody is then purified by Sephadex G-50 column using 0.15 M NaCl as eluent.

EXAMPLE 5

Non-site-selective labeling of $F_{ab'}$ fragments with technetium-99m.

A mixture of the $F_{ab'}$ fragment and about 25 fold excess of the ligand shown in Scheme 2 is incubated in 25 mM phosphate buffer, pH 7.4 at room temperature for about 30 minutes. Excess ligand and other low molecular weight impurities are quickly removed by Sephadex G-25 column using the same buffer. Thereafter, the conjugated $F_{ab'}$ solution was treated with 15 μL ^{99m}Tc-saccharic acid and the mixture is incubated at 37°C for about 30 minutes. The technetium labeled antibody is then purified by Sephadex G-

25 column using the same buffer.

In order to image biological receptors, a preparation of the present invention using either whole, fragmented, or recombinant anti-idiotypic antibodies or a recombinant fragment thereof is administered to the patient, for example, in the form of an injectable liquid. By means of suitable detectors, e.g., a gamma camera, images can be obtained by recording the emitted radiation of the organ or the pathological process in which the labeled anti-idiotypic antibody has been incorporated, which in the present case is biological receptors.

The anti-idiotypic antibody of the present invention or a fragment or recombinant derivative thereof prepared as described above provides a means of in vivo diagnostic imaging of receptors which provides many advantages over prior known procedures which involve cellular secretions.

After the anti-idiotypic antibody is prepared and labeled according to one of the procedures described, the composition is used with a pharmaceutically acceptable carrier in a method of performing a diagnostic imaging procedure using a gamma camera or like device which involves injecting or administering to a warm-blooded animal an effective amount of the present invention and then exposing the warm-blooded animal to an imaging procedure as described above, thereby imaging at least a portion of the body of the warm-blooded animal.

Pharmaceutically acceptable carriers include those that are suitable for injection such as aqueous buffer solutions, e.g., tris(hydroxymethyl)aminomethane (and its salts), phosphate, citrate, bicarbonate, etc., sterile

water for injection, physiological saline, and balanced ionic solutions containing chloride and/or bicarbonate salts of normal blood plasma cations such as Ca^{2+}, Na^{+}, K^{+} and Mg^{2+}. Other buffer solutions are described in *Remington's Practice of Pharmacy*, Eleventh Edition, for example on page 170. The carriers may contain a chelating agent, e.g., a small amount of ethylenediaminetetraacetic acid, calcium disodium salt, or other pharmaceutically acceptable chelating agents.

The concentration of the labeled anti-idiotypic antibodies in the pharmaceutically acceptable carrier, for example an aqueous medium, varies with the particular field of use. A sufficient amount is present in the pharmaceutically acceptable carrier in this particular case when satisfactory visualization of the receptors is achievable.

The composition is administered to the warm-blooded animal so that the composition remains in the living animal body for about 6 to 7 hours, although shorter and longer residence periods are normally acceptable.

The labeled anti-idiotypic antibodies may be used in the usual way in imaging procedures. For example, with the present invention when imaging biological receptors, a sufficient amount of the labeled anti-idiotypic antibody must be intravenously administered to the warm-blooded animal to provide adequate visualization; the animal or a portion thereof is then scanned with a suitable imaging machine such as a gamma camera.

After consideration of the above specification, it will be appreciated that many improvements and modifications in the details may be made without departing

from the spirit and scope of the invention. It is to be understood, therefore, that the invention is in no way limited, except as defined by the appended claims.

CLAIMS:

1. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, or recombinant anti-idiotypic antibody labeled with a chelate for intravenous injection into an animal to produce reliable visual imaging of biological receptors.

2. A method of performing a diagnostic procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, or recombinant anti-idiotypic antibody labeled with a chelate to allow for visual imaging of biological receptors.

3. A whole, fragmented, or recombinant anti-idiotypic antibody labeled with a chelate for intravenous injection into a warm-blooded animal to produce reliable visual imaging of biological receptors.

4. The whole, fragmented, or recombinant anti-idiotypic antibody of claims 1, 2, or 3 wherein said anti-idiotypic antibody is labeled with a chelate for injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

5. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment labeled with a radionuclide bound chelate capable of intravenous injection into an animal to produce reliable visual imaging of biological receptors.

6. A method of performing a diagnostic

procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented recombinant anti-idiotypic antibody or recombinant fragment labeled with a radionuclide bound chelate to allow for visual imaging of biological receptors.

7. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment labeled with a radionuclide bound chelate for intravenous injection into a warm-blooded animal to produce reliable visual imaging of biological receptors.

8. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 5, 6, or 7 wherein said anti-idiotypic antibody is labeled with a radionuclide bound chelate for injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

9. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled with Tc-99m for intravenous injection into an animal to produce reliable visual imaging of digoxin receptors located in myocardium tissue.

10. A method of performing a diagnostic procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled with Tc-99m to allow for visual imaging of digoxin receptors located in myocardium tissue.

11. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled with Tc-99m for intravenous injection into a warm-blooded animal to produce reliable visual imaging of digoxin receptors located in myocardium tissue.

12. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 9, 10, or 11 wherein said anti-idiotypic antibody is radiolabeled with Tc-99m for injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

13. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with digoxin receptor specificity labeled with a chelate for intravenous injection into an animal to produce reliable visual imaging of digoxin receptors located in myocardium tissue.

14. A method of performing a diagnostic procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with digoxin receptor specificity labeled with a chelate to allow for visual imaging of digoxin receptors located in myocardium tissue.

15. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with digoxin specificity labeled with a chelate for intravenous injection into a warm-blooded animal to produce reliable visual imaging of digoxin receptors located in myocardium tissue.

16. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 13, 14, or 15 wherein said anti-idiotypic antibody labeled with a chelate for injection into a warm-blooded animal to produce reliable visual imaging of digoxin receptors located in myocardium tissue within two and one half hours post-injection.

17. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment labeled with fluorine atoms for intravenous injection into an animal to produce reliable visual imaging of biological receptors.

18. A method of performing a diagnostic procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment labeled with fluorine atoms to allow for visual imaging of biological receptors.

19. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment labeled with a fluorinated chelate for intravenous injection into a warm-blooded animal to produce reliable visual imaging of biological receptors.

20. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 17, 18, or 19 wherein said anti-idiotypic antibody labeled with a fluorinated chelate for injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

21. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled for intravenous injection into an animal to produce reliable visual imaging of biological receptors.

22. A method of performing a diagnostic procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled to allow for visual imaging of biological receptors.

23. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment radiolabeled for intravenous injection into a warm-blooded animal to produce reliable visual imaging of biological receptors.

24. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 21, 22, or 23 wherein said anti-idiotypic antibody radiolabeled is capable of injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

25. A diagnostic composition suitable for administration to a warm-blooded animal comprising a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with receptor specificity labeled with a paramagnetic metal chelate for intravenous injection into an animal to produce reliable visual imaging of biological receptors.

26. A method of performing a diagnostic

procedure, which comprises administering to a warm-blooded animal an imaging-effective amount of a whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with receptor specificity labeled with a paramagnetic metal chelate to allow for visual imaging of biological receptors.

27. A whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment with receptor specificity labeled with a paramagnetic metal chelate capable of intravenous injection into a warm-blooded animal to produce reliable visual imaging of biological receptors.

28. The whole, fragmented, recombinant anti-idiotypic antibody or recombinant fragment of claims 25, 26, or 27 wherein said anti-idiotypic antibody labeled with a paramagnetic metal chelate is capable of injection into a warm-blooded animal to produce reliable visual imaging of biological receptors within two and one half hours post-injection.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US92/05500

A. CLASSIFICATION OF SUBJECT MATTER
IPC(5) :A61K 49/02, 37/04; C07K 13/00, 15/28
US CL :424/85.8, 1.1, 9; 530/387.2, 391.3
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 424/85.8, 1.1, 9; 530/387.2, 391.3, 391.7; 435/7.21, 7.23

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

APS, DIALOG
search terms: anti-idiotypic antibodies, imaging

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X Y	Cancer Research (Suppl.), Volume 50, issued 01 February 1990, B.A. Parker et al, "Radioimmunotherapy of Human B-Cell Lymphoma with 90Y-conjugated Antiidiotype Monoclonal Antibody" pages 1022s-1028s, see abstract and pages 1023s, 1024s, and 1026s.	1-8, 21-24 9-20, 25-28
Y	Immunological Reviews, No. 94, issued 1986, B.F. Erlanger et al, "Auto-Anti-Idiotypy: A Basis for Autoimmunity and a Strategy for Anti-Receptor Antibodies", pages 23-37, see pages 33 and 35.	9-16
Y	US, A, 4,606,855 (Deutsch et al) 19 August 1986, see columns 3 and 4.	9-16
Y	D.M. Goldenberg, "Cancer Imaging with Radiolabeled Antibodies" published 1990 by Kluwer Academic Publishers, see pages 233-244, especially page 233.	9-16
Y	Magnetic Resonance in Medicine, Vol. 5, issued 1987, Shimizu et al, "Tumor Imaging with Anti-CEA Antibody Labeled 19F Emulsion", pages 290-295, see pages 292-293	17-20

[X] Further documents are listed in the continuation of Box C. [ ] See patent family annex.

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Date of the actual completion of the international search	Date of mailing of the international search report
24 August 1992	28 AUG 1992
Name and mailing address of the ISA/ Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. NOT APPLICABLE	Authorized officer LORA M. GREEN Telephone No. (703) 308-0196

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INTERNATIONAL SEARCH REPORT

International application No.
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C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US, A, 4,659,839 (Nicolotti et al) 21 April 1987, see column 1.	25-28